Alterations in the 5 ’untranslated region of the *EPSPS* gene influence *EPSPS* over-expression in glyphosate-resistant *Eleusine indica*

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Summary The herbicide glyphosate inhibits the enzyme 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS). Over-expression of the *EPSPS* gene is one of the molecular mechanisms conferring glyphosate resistance in weeds, but the transcriptional regulation of this gene is poorly understood. The *EPSPS* gene was found to be significantly up-regulated following glyphosate treatment in a glyphosate- resistant *Eleusine indica* population from South China. To further investigate the regulation of *EPSPS* over-expression, the promoter of the *EPSPS* gene from this *E. indica* population was cloned and analysed. Two upstream regulatory sequences, Epro-S (862 bp) and Epro-R (877 bp) of *EPSPS* were obtained respectively from glyphosate-susceptible and -resistant *E. indica* plants by HiTAIL-PCR. The Epro-S and Epro-R sequences were 99% homologous, except for the two insertions (3 bp and12 bp) in the Epro-R sequence. The 12-base insertion of the Epro-R sequence was located in the 5′-UTR-Py-rich stretch element. The promoter activity tests showed that the 12-base insertion resulted in significant enhancement of the Epro-R promoter activity, whereas the 3-base insertion had little effect on Epro-R promoter activity.

In conclusion, alterations in the 5′-UTR-Py-rich stretch element of *EPSPS* are responsible for glyphosate induced *EPSPS* over-expression. Therefore, *EPSPS* transcriptional regulation conferred glyphosate resistance in this *E. indica* population.

Keywords *Eleusine indica*, EPSPS, promoter, 5′-UTR-Py-rich stretch element, insertion.